Occurrence of the plasmid-borne mcr-1 colistin resistance gene in ESBL-producing Enterobacteriacae in river water and imported vegetable samples in Switzerland

Zurfluh, Katrin ; Poirel, Laurent ; Nordmann, Patrice ; Nüesch-Inderbinen, Magdalena ; Hächler, Herbert ; Stephan, Roger

DOI: https://doi.org/10.1128/AAC.00066-16

Antimicrobial Agents and Chemotherapy – New Data Letter

Occurrence of the plasmid-borne *mcr-1* colistin resistance gene in ESBL-producing Enterobacteriacae in river water and imported vegetable samples in Switzerland

Katrin Zurfuh[1], Laurent Poirel[2], Patrice Nordmann[2,3], Magdalena Nüesch-Inderbinen[1], Herbert Hächler[1], Roger Stephan[1*]

[1]*Institute for Food Safety and Hygiene, Vetsuisse Faculty University of Zurich Winterthurerstrasse 272, 8057 Zurich, Switzerland*

[2]*Emerging Antibiotic Resistance Unit, Medical and Molecular Microbiology, Department of Medicine, Faculty of Science, University of Fribourg*

[3]*HFR - Hôpital Cantonal, Fribourg, Switzerland*

[*]Corresponding author

Roger Stephan, Institute for Food Safety and Hygiene, Vetsuisse Faculty University of Zurich, Winterthurerstr. 272, CH-8057 Zurich, Switzerland, Phone 0041-44-6358651, fax 0041-44-6358908, e-mail stephanr@fsafety.uzh.ch

The recent identification of Enterobacteriaceae harbouring the plasmid-mediated transferable colistin resistance *mcr-1* gene is of great concern to public health (1-4). Here, we report on the occurrence of *mcr-1* harbouring extended-spectrum ß-lactamase (ESBL)-producing Enterobacteriaceae from river water in Switzerland and ready-to-eat imported vegetables.

For this study, 74 ESBL-producing *Enterobacteriaceae* isolated from 21 rivers and lakes sampled in 2012 in Switzerland (5) and 60 ESBL-producing *Enterobacteriaceae* isolated from 42 imported vegetable samples (11 from the Dominican Republic, 13 from India, 11 from Thailand and 8 from Vietnam) (6) were screened by PCR for the presence of the *mcr-1* gene.

The gene was detected in one out of 74 water strains (an isolate from the river "Birs") and 2 out of 60 vegetable strains (products from Thailand and Vietnam) and sequencing of the amplicons showed a 100% identity with the published *mcr-1* sequence (1). The colistin resistance was transferable by transformation experiments into *E. coli* DH5-alpha. All strains were *Escherichia coli* and belonged to different multi locus sequence types (MLST), harboured different bla_{ESBL} genes and showing a multiresistance phenotype (Table 1). The diversity of ESBL genes and MLST types identified among *mcr-1*-positive isolates suggests that the *mcr-1* gene might be carried on different plasmids.

The spread of *mcr-1* harbouring ESBL-producing Enterobacteriaceae in surface water suggest environmental contamination. Appropriate measures urgently need to be enforced in order to reduce the anthropogenic burden of antibiotic resistance in the environment, such as the judicious use of antibiotics in human and veterinary medicine as well as in agriculture. In addition, improvement of water status is of major concern. New strategies for the treatment of wastewaters, e.g., the use of sand filters or more-stringent chlorine disinfection, need to be taken into consideration to prevent resistant bacteria from being released into the aquatic environment. Moreover, these data show that the international production and trade of fresh vegetables constitute a possible route for the spread of antibiotic-resistant, and particularly

colistin-resistant Enterobacteriaceae.

References

1. **Liu YY, Wang Y, Walsh TR, Yi LX, Zhang R, Spencer J, Doi Y, Tian G, Dong B, Huang X, Yu LF, Gu D, Ren H, Chen X, Lv L, He D, Zhou H, Liang Z, Liu JH, Shen J.** 2015. Emergence of plasmid-mediated colistin resistance mechanism MCR-1 in animals and human beings in China: a microbiological and molecular biological study. Lancet Infect. Dis. (published online November 18.) http://dx.doi.org/10.1016/S1473-3099(15)00424-7
2. **Arcilla MS, van Hattem JM, Matamoros S, Melles DC, Penders J, de Jong MD, Schultsz C.** 2015. Dissemination of the *mcr-1* colistin resistance gene. Lancet Infect. Dis.(published online December 17.) http://dx.doi.org/10.1016/S1473-3099(15)00541-1
3. **Granier SA, Marault M, Millemann Y, den Bakker HC, Nightingale KK, Bugarel M, Ison SA, Morgan Scott HM, Loneragan GH.** 2015. Dissemination of the *mcr-1* colistin resistance gene. Lancet Infect. Dis. (published online December 17.) http://dx.doi.org/10.1016/S1473-3099(15)00538-1
4. **Hasman H, Hammerum AM, Hansen F, Hendriksen RS, Olesen B, Agersø Y, Zankari E, Leekitcharoenphon P, Stegger M, Kaas RS, Cavaco LM, Hansen DS, Aarestrup FM, Skov RL.** 2015. Detection of *mcr-1* encoding plasmid-mediated colistin-resistant *Escherichia coli* isolates from human bloodstream infection and imported chicken meat, Denmark 2015. Euro Surveill. 10;20(49). doi: 10.2807/15607917.ES.2015.20.49.30085.
5. **Zurfluh K, Hächler H, Nüesch-Inderbinen M, Stephan R.** 2013. Characteristics of extended-spectrum beta-lactamase (ESBL)- and carbapenemase-producing Enterobacteriaceae isolated from rivers and lakes in Switzerland. Appl. Environ. Microbiol. 79, 3021-3026.

6. **Zurfluh K, Nüesch-Inderbinen M, Morach M, Zihler Berner A, Hächler H, Stephan R.** 2015. Extended-spectrum ß-lactamase-producing-Enterobacteriaceae in vegetables imported from the Dominican Republic, India, Thailand and Vietnam. Appl. Environ. Microbiol. **81**: 3115–3120.

Table 1. Characteristics and resistance profiles of ESBL-producing and *mcr-1*-positive Enterobacteriaceae

	Strain OW3E1	Strain H226B	Strain 2SK1
Sample type	River water sampled in Switzerland	Cha-om imported from Thailand	Basil leaves imported from Vietnam
Year	2012	2014	2014
Species	*E. coli*	*E. coli*	*E. coli*
Phylogroup	B1	A	B1
Sequence type	ST359	ST167	ST4683
bla_{ESBL}	SHV-12	CTX-M55	CTX-M-65
MIC for colistin (μg/mL)	6	6	6
AM	R	R	R
AMC	S	S	R
CF	R	R	R
CTX	R	R	R
CIP	R	R	R
GM	S	R	R
TE	R	R	R
S	R	R	R
C	R	S	R
K	S	R	S
NA	R	R	R
SMZ	R	R	R
TMP	R	S	R

ampicillin(AM), amoxicillin-clavulanic acid (AMC), cephalothin (CF), cefotaxime (CTX), ciprofloxacin (CIP), gentamicin (GM), tetracycline (TE), streptomycin (S), chloramphenicol (C), kanamycin (K), nalidixic acid (NA), sulfamethoxazole (SMZ), and trimethoprim (TMP)
R: resistant, S: susceptible